Report Documentation Page

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
FEB 2010		**00-00-2010 to 00-00-2010**

4. TITLE AND SUBTITLE	5a. CONTRACT NUMBER
1.3 Mb de novo deletion in chromosome band 3q29 associated with normal intelligence in a child	5b. GRANT NUMBER
	5c. PROGRAM ELEMENT NUMBER
6. AUTHOR(S)	5d. PROJECT NUMBER
	5e. TASK NUMBER
	5f. WORK UNIT NUMBER
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) **National Naval Medical Center,Department of Pediatrics,8901 Wisconsin Avenue,Bethesda,MD,20889-5600**	8. PERFORMING ORGANIZATION REPORT NUMBER
9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)	10. SPONSOR/MONITOR'S ACRONYM(S)
	11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION/AVAILABILITY STATEMENT
Approved for public release; distribution unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

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15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			
unclassified	**unclassified**	**unclassified**	**Same as Report (SAR)**	**5**	

European Journal of Medical Genetics 53 (2010) 415–418

Contents lists available at ScienceDirect

European Journal of Medical Genetics

journal homepage: http://www.elsevier.com/locate/ejmg

Chromosomal imbalance letter

1.3 Mb *de novo* deletion in chromosome band 3q29 associated with normal intelligence in a child

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ARTICLE INFO

Article history:
Received 4 February 2010
Accepted 30 August 2010
Available online 9 September 2010

Keywords:
Deletion 3q29
IQ
Autism
Array CGH analysis

ABSTRACT

We report on a 6 and 9/12 year-old male patient with a *de novo* chromosome 3q29 microdeletion identified by BAC array comparative genomic hybridization assay (aCGH), with accompanying normal 46,XY high-resolution chromosome analysis. The patient has language-based learning disabilities and behavioral features consistent with diagnoses of autism and attention deficit hyperactivity disorder (ADHD) of the inattentive type. He also displays some other features previously associated with chromosome 3q29 microdeletion such as an elongated face, long fingers, and joint laxity. Most notably our patient, per formal IQ testing, was not found to have frank mental retardation as has been previously reported among patients with chromosome 3q29 terminal deletion, but rather our patient has demonstrated an average full-scale IQ result. Our report further expands the phenotypic spectrum of the rare chromosome 3q29 microdeletion syndrome to include the possibility of normal intelligence as corroborated by formal, longitudinal psycho-educational testing.

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1. Methods of detection

An aCGH was performed using the ClariSure™ (Quest Diagnostics Nichols Institute, San Juan Capistrano, CA) 1 Mb array built from commercially available BAC clones (IntegraGen, Invitrogen, and Open Biosystems). The BAC clones were printed in duplicate onto aminosilane coated glass slides (Corning) using the BioRobotics Microgrid II Printer (Genomic Solutions) to generate 2 arrays per slide. Dual color aCGH was performed using Cy3-dCTP and Cy5-dCTP (GE Healthcare) and Human Cot-1 DNA (Invitrogen). The slides were scanned on the GenePix 4000B Scanner using Genepix 6.0 software. The dual color aCGH was analyzed using Quest Diagnostics independently developed and validated oneClickCGH® software (Infoquant). The aCGH was performed in duplicate with 3176 informative BAC probes targeting multiple loci across all chromosomes. A threshold of ±4 SD was used as a reference for discrimination of genomic alterations [9].

2. Chromosomal anomaly

This aCGH assay exhibited ratio plots consistent with a deletion of at least 1.3 Mb from the distal long arm of chromosome 3 at band 3q29, involving at least the region encompassed by nucleotides 3:197,256,140–198,570,020 (UCSC 2006 hg18). Specifically, the three most distal adjacent BAC clones on this array (CTD-3185F8, CTD-2334B5, RP11-233N20) were deleted.

3. Method of confirmation

This deletion was confirmed by a supplemental Fluorescent In-Situ Hybridization (FISH) assay using the Vysis/Abbott Molecular 3q subtelomere probe on metaphase and interphase cells.

4. Clinical description

Our patient was a 6 and 9/12 year-old male who is the fourth of five children and was born at term gestation to healthy non-consanguineous parents. Prenatal history for the patient was only notable for maternal vaginal bleeding at 10 weeks gestation that resolved with bed rest. There was no reported drug, alcohol, or

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1769-7212/$ – see front matter Published by Elsevier Masson SAS.
doi:10.1016/j.ejmg.2010.08.009

other chemical exposure. The patient was born vaginally at 38 + 6 weeks gestation and weighed 2583 g (6th centile for term infant male) with length of 49.5 cm (43rd cent.) and head circumference of 33 cm (10th cent.). His newborn course was unremarkable. Pediatric history was notable for symptoms of constipation and malabsorption with pancreatic insufficiency (on enzyme replacement) and poor weight gain during the first year of life. He has also been treated for gastro-esophageal reflux with eosinophilic esophagitis. He was diagnosed with autism at age 3.5 years, attention deficit hyperactivity disorder predominantly inattentive type at age 6 years 11 months, and speech delay. He is treated for seasonal allergies. He has had normal renal ultrasound, brain magnetic resonance imaging, and auditory testing as well as normal lab testing to include sweat chloride testing, fragile X DNA testing, and celiac screening.

Developmentally the proband sat independently at 6.5 months, crawled at 10 months, spoke his first word(s) at 15 months, walked at 16 months, and was evaluated for speech and pervasive developmental delay at approximately 3 years of age. He previously received speech therapy through the public school system and currently has 25 h weekly special education services and 2 h monthly occupational therapy services. His receptive language has tested in the normal range with much weaker expressive language skills often testing <5th cent. He started initially in a pre-school program for autistic children after diagnosis at 3.5 years of age, transitioned to public mainstream kindergarten at 5 years of age and is currently in the first grade with "Good" to "Satisfactory" reported performance in all academic areas.

Stanford-Binet IQ testing performed at 5 years of age showed a full scale IQ of 87 (19th cent., CI 83–91, low average), nonverbal IQ of 99 (47th cent., CI 93–105, average) and verbal IQ of 77 (6th cent., CI 72–84, borderline). Repeat IQ testing was performed at age 6 years 11 months utilizing the Wechsler Intelligence Scale for children (4th ed.). Composite IQ scores ranged from 74 (working memory, 4th cent., CI 68–84, borderline) to 96 (verbal comprehension, 39th cent., CI 89–103, average) with full scale IQ of 84 (14th cent., CI 80–89, low average). Because of significant composite score variability a more appropriate General Ability Index (GAI) of 94 (34th cent., average) reflects overall average reasoning abilities. Areas of strengths were noted in associative reasoning, vocabulary, and spatial organization. Areas of weakness were noted in verbal formulation and working memory tasks. Academic achievement, as measured using the Wide Range Achievement Test – revision 4 (WRAT 4) at age 6 years 11 months, was also highly variable and ranged from a standard age-based score of 80 (9th cent., math computation, K4 grade level) to 118 (88th cent., word reading, grade 2.8 level). Areas of strengths were individual word reading and sentence comprehension. Areas of weakness were math computation and spelling.

Our patient's mother was 37 years old at the time of his birth, and she is of French, Italian, and German descent. She is currently a homemaker but previously worked outside the house in a professional position. Her past medical history is notable for a right sided aortic arch without dextrocardia, prompting a fetal echocardiogram for our patient that was normal. His father is of German and Scottish background and was 43 years old at the time of our patient's birth. He works outside the home in a professional position. Both parents are college educated, including two master degrees earned by his mother. There is no family history of learning difficulties, birth defects or mental retardation. The patient has three older siblings and one younger sibling reportedly in good health with normal intelligence. The younger sibling has had chromosomal and subtelomeric FISH studies per parental request which were reported as normal, as were the parental chromosomal and subtelomeric FISH studies. Review of family photos and photos of the patient over time were remarkable only for the patient's elongated face, more prominent ears, and higher nasal bridge when compared to his siblings.

Physical examination revealed a relatively tall and slender child with weight 21.6 kg (45th cent. for age), height of 125 cm (85th cent. for age), and head circumference of 50.5 cm (25th cent. for age). He was described as "clumsy" at times by his mother, but no gait ataxia was noted. He had very mildly decreased general tone with notable hypermobility of his fingers and hands. He had an elongated face with downslanting palpebral fissures, and normally set but somewhat prominent ears (Fig. 1). He had a high nasal bridge with prominent tip, short but well-delineated philtrum, and normal palate and teeth. He has elongated fingers with normal palmar creases and mild fifth finger clinodactyly bilaterally (Fig. 1). Pes planus was noted bilaterally as well as laterally deviated second toes (Fig. 1) which was reported to be a familial trait.

5. Discussion

Literature review on a total of 22 cases identified with similar chromosome 3q29 microdeletions as our patient's deletion reveals variable phenotypes associated with the 3q29 microdeletion

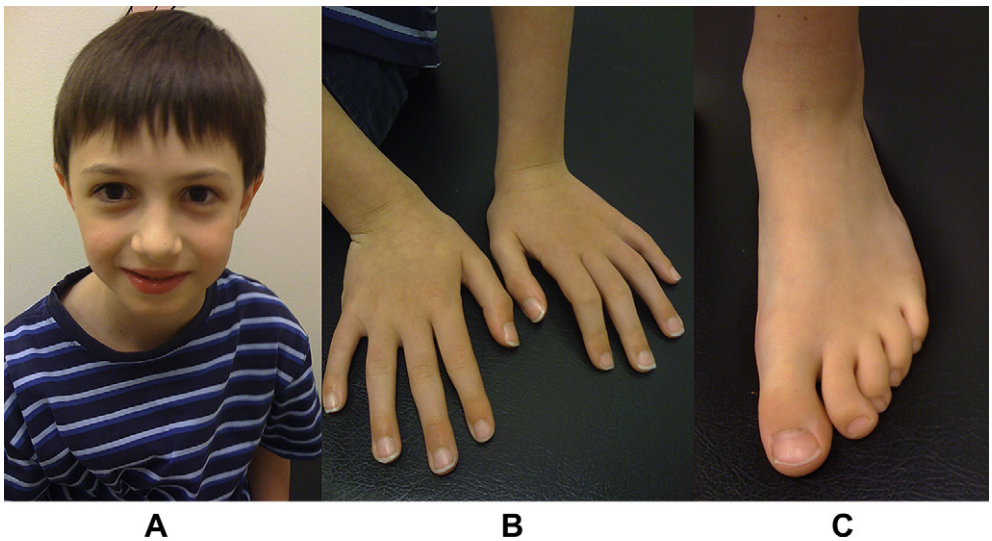

Fig. 1. (A) Patient at 6 years 9/12 months of age. (B) Long fingers and mild 5th finger clinodactyly. (C) Laterally deviated 2nd toe (present bilaterally).

Table 1
Select clinical features reported in literature for patients with 3q29 microdeletion syndrome in order of decreasing prevalence (modified from Tyshchenko et al. [7], and Ballif et al. [1]).

Clinical Feature	This patient	Previously reported[a]	Total
Developmental delay/mental retardation		21/22	21/23
High nasal bridge	+	13/22	14/23
Microcephaly		11/22	11/23
Short philtrum	+	10/22	11/23
Delayed walking		9/20	9/21
Large posteriorly rotated ears	+	7/22	8/23
Ataxic gait		5/20	5/21
Autistic features	+	4/20	5/21
Digit clinodactyly	+	4/22	5/23
Long narrow face	+	3/22	4/23
Cardiac defect		3/22	3/23
Chest wall deformities		3/22	3/23
Recurrent middle ear infections		3/22	3/23
High arched palate		2/22	2/23
Widely spaced teeth		2/20	2/21
Genital abnormalities		2/21	2/22
Nasal Voice		2/22	2/23
Ligamentous laxity	+	1/22	2/23
Macrocephaly		1/22	1/23
Cleft lip/palate		1/22	1/23
Abnormal skin pigmentation		1/22	1/23
Horseshoe kidney		1/22	1/23
Six lumbar vertebrae		1/22	1/23
Vascular anomalies		1/22	1/23
Ocular abnormalities		1/22	1/23

[a] Not all features could be assessed due to age of patient or not reported on all patients.

syndrome (Table 1) [3–5]. Mild to moderate mental retardation was identified in all previously identified individuals with the possible exception of one infant/father pair [4]. We herein identify an additional patient with this chromosome deletion who exhibits several previously documented physical traits but has a formally documented overall low average to average intelligence quotient.

It is important to point out that since our patient also exhibits traits consistent with autism and ADHD, his IQ scores and academic achievement may reflect these diagnoses in addition to chromosome 3 microdeletion syndrome. Autism has a known association in persons with 3q microdeletions, with four of the previously reported twenty-two patients with this syndrome also displaying autistic features [7]. Despite this developmental challenge, our patient continues in an age appropriate classroom with an individualized education program in place that allows for additional schooling assistance. He also received and likely benefitted from early speech therapy intervention. He currently receives and presumptively benefits from additional special education services. The otherwise relative normalcy of his functioning in a mainstream school setting is very encouraging when one considers the more serious developmental outcome usually associated with chromosome 3q subtelomeric deletions and autism, as well as the learning difficulties commonly associated with ADHD.

Other features found in our patient and consistent with previously reported patient features include his elongated face, relatively high nasal bridge with prominent tip, somewhat prominent ears, mild joint laxity (hands and fingers), short philtrum, and fifth finger clinodactyly. He does not demonstrate microcephaly, ataxic gait, or chest wall deformity as has been previously reported with chromosome 3q deletions (Table 1).

Our patient's 3q29 *de novo* terminal deletion is at least 1.3 Mb in size and encompasses at least 19 genes. The breakpoints fall within the typically reported region of the other affected patients [2–4] (Fig. 2). Included within the deleted region notably are genes for DLG1 and PAK2, autosomal homologues of known mental retardation X-linked genes PAK3 and DLG3. Previous reports on patients with 3q29 microdeletions that include genes for DLG1 and PAK2 have postulated on these genes' contribution to impaired neurosynaptic development and ensuing mental retardation [2,10].

Given the rarity of patients with this microdeletion syndrome as well as the relatively smaller patient population that presents for chromosomal analysis, there is an ascertainment bias for finding chromosomal abnormalities in selected patients who present with mental retardation or other developmental disabilities such as autism. At least one study of seven individuals subsequently identified as having terminal chromosome 3q deletions were initially screened based upon the presence of mental retardation, developmental delay, or multiple congenital anomalies [1]. The use of subtelomeric deletion screening and/or aCGH to investigate such patients, particularly if dysmorphic features are also present, has become standard laboratory investigation [6,8]. At least twenty of the previously described twenty-two patients with this syndrome have been described to have at least mild mental retardation. More recently a 6-month old infant and father, both with 3q29 microdeletion and cardiac defects, were described as developmentally normal which suggests at least the possibility of normal intelligence [4]. We believe our study is complementary to these encouraging findings and additionally includes results of comprehensive formal IQ testing in an older child. As evidenced in our

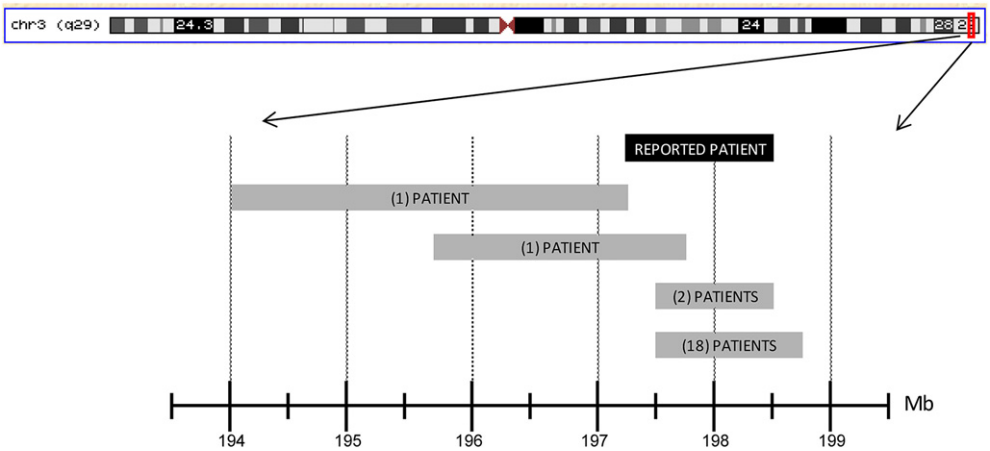

Fig. 2. Schematic comparison of previously reported twenty-two patients with 3q29 microdeletions similar to our patient. Deleted regions are approximated. Figure was partially generated by using the UCSC Genome Browser (hg18).

patient, frank mental retardation may not be a necessary feature when describing the 3q29 microdeletion syndrome phenotype.

In conclusion, we report on a child with chromosome 3q29 microdeletion to expand further on the phenotype of this disorder, and to underscore that mental retardation need not be present, thereby giving a more favorable developmental prognosis for this patient population.

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